

This Page Is Inserted by IFW Operations
and is not a part of the Official Record

BEST AVAILABLE IMAGES

Defective images within this document are accurate representations of the original documents submitted by the applicant.

Defects in the images may include (but are not limited to):

- BLACK BORDERS
- TEXT CUT OFF AT TOP, BOTTOM OR SIDES
- FADED TEXT
- ILLEGIBLE TEXT
- SKEWED/SLANTED IMAGES
- COLORED PHOTOS
- BLACK OR VERY BLACK AND WHITE DARK PHOTOS
- GRAY SCALE DOCUMENTS

IMAGES ARE BEST AVAILABLE COPY.

**As rescanning documents *will not* correct images,
please do not report the images to the
Image Problem Mailbox.**



Attorney's Docket No.: 12557-004001

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicant : Andrew Kloeck et al. Art Unit : 1652
Serial No. : 10/098,602 Examiner : S. Swope
Filed : March 15, 2002
Title : NEMATODE GS-LIKE SEQUENCES

DECLARATION OF ANDREW P. KLOECK UNDER 37 C.F.R. §1.132

1. I, Andrew P. Kloeck, am Senior Director at Divergence, Inc. I received a Ph.D. in Molecular Genetics from Washington University (St. Louis, MO) in 1995 and a B.S in Genetics from Western Kentucky University (Bowling Green, KY) in 1991. I am an inventor on the above-captioned patent application.

2. The *Meloidogyne incognita* protein identified in the above-referenced patent application as SEQ ID NO:2 was identified as a glutamine synthetase (GS)-like protein based, in part, on Pfam analysis.

3. Pfam is a curated database of protein domain families and associated analytical tools. The database and the analytical tools associated with the database are designed to provide more accurate assessment of protein function than can be achieved by simple pair-wise sequence comparisons such as BLAST analysis. Pfam analysis involves comparing a test sequence, here SEQ ID NO:2, to Hidden Markov Model (HMM) descriptions of more than 1800 protein domain families, each of which has multiple representative members. Pfam analysis assigns an S-score for each comparison between the test protein and the HMM of the protein domain family. This S-score is a

CERTIFICATE OF MAILING BY FIRST CLASS MAIL

I hereby certify under 37 CFR §1.8(a) that this correspondence is being deposited with the United States Postal Service as first class mail with sufficient postage on the date indicated below and is addressed to the Commissioner for Patents, P.O. Box 1450, Alexandria, VA 22313-1450.

Date of Deposit

July 15, 2004

Signature

Carrie A. Amonte

Typed or Printed Name of Person Signing Certificate

Carrie A. Amonte



measure of the relatedness of the test protein to the particular protein domain family. The S-score has an associated "e-value". The e-value is a measure of the odds that the particular S-score for the test protein (or an even better S-score) could arise by chance. Put another way, the e-value is the number of hits that would be expected to have a score equal or better than this S-score by chance alone. Thus, the lower the e-value, the lower the likelihood the match between the test protein and the protein domain family is a chance match and the greater the likelihood that the test protein is a member of the protein domain family. Furthermore, the manual curators of individual Pfam protein models set a score threshold called the "gathering threshold" (GA cutoff) which is considered to be trustworthy cutoff above which effectively zero false positives get through.

4. The *M. incognita* gene encoding SEQ ID NO:2 was identified in a search for *M. incognita* ESTs with sequence similarity to the *Mycobacterium tuberculosis* glutamine synthetase gene glnA4 (GenBank® Accession No. F70885) as described in pages 18 to 19 of the specification.

5. When the *M. tuberculosis* glutamine synthetase gene glnA4 (GenBank® Accession No. F70885) is subjected to Pfam analysis, the domain model sequence with the best e-value (i.e., lowest) is the glutamine synthetase domain model sequence. Pfam analysis assigns an e-value of $4.2e^{-89}$ to this comparison. This value indicates that the match is highly unlikely to occur by chance.

6. Pfam analysis of SEQ ID NO:2 revealed that the domain model sequence with the best e-value was the glutamine synthetase domain model sequence, which was assigned an e-value of $1.7e^{-77}$. This value indicates that the match is highly unlikely to occur by chance. This Pfam analysis strongly supports the conclusion that SEQ ID NO:2 is a glutamine synthetase-like enzyme. The Pfam analysis indicates that the predicted glutamine synthetase domain of SEQ ID NO:2 extends from amino acid 115 to 375.

7. A BLAST analysis that compares SEQ ID NO:2 to protein sequences currently in the GenBank® database reveals that nearly all of the proteins that have sequence significantly similar to

SEQ ID NO:2 are identified as glutamine synthetases. This analysis supports the conclusion that SEQ ID NO:2 is a glutamine synthetase-like enzyme. The results of this BLAST analysis is shown below.

Sequences producing significant alignments:	(bits) Value
gil13472698 ref NP_104265.1	glutamine synthetase [Mesorhiz... 384 e-105
gi23500469 ref NP_699909.1	glutamine synthetase family pr... 366 e-100
gi15965966 ref NP_386319.1	CONSERVED HYPOTHETICAL PROTEIN... 361 2e-98
gi15889423 ref NP_355104.1	AGR_C_3883p [Agrobacterium tum... 357 4e-97
gi17988899 ref NP_541532.1	GLUTAMINE SYNTHETASE [Brucella... 350 5e-95
gi45917208 ref ZP_00196362.2	COG0174: Glutamine synthetas... 336 6e-91
gi15609997 ref NP_217376.1	GlnA4 [Mycobacterium tuberculo... 303 7e-81
gi21220110 ref NP_625889.1	putative glutamine synthetase ... 302 1e-80
gi22958785 ref ZP_00006449.1	COG0174: Glutamine synthetas... 301 2e-80
gi41409029 ref NP_961865.1	GlnA4 [Mycobacterium avium sub... 294 3e-78
gi29833267 ref NP_827901.1	putative glutamine synthetase ... 292 9e-78
gi46362694 ref ZP_00225545.1	COG0174: Glutamine synthetas... 278 2e-73
gi46106195 ref ZP_00199871.1	COG0174: Glutamine synthetas... 273 6e-72
gi15865464 emb CAC81335.1	gamma-glutamylisopropylamide sy... 218 3e-55
gi48856911 ref ZP_00311068.1	COG0174: Glutamine synthetas... 211 3e-53
gi24461668 gb AAN62237.1	putative glutamine-synthetase [P... 208 2e-52
gi46915963 emb CAG22734.1	hypothetical protein [Photobact... 182 2e-44
gi28869125 ref NP_791744.1	glutamine synthetase [Pseudomo... 178 3e-43
gi46199267 ref YP_004934.1	glutamine synthetase [Thermus ... 177 6e-43
gi48477605 ref YP_023311.1	glutamine synthetase [Picrophi... 176 1e-42
gi46311445 ref ZP_00212051.1	COG0174: Glutamine synthetas... 172 1e-41
gi46321429 ref ZP_00221806.1	COG0174: Glutamine synthetas... 172 2e-41
gi15616429 ref NP_244734.1	glutamine synthetase [Bacillus... 171 5e-41
gi22972120 ref ZP_00019019.1	hypothetical protein [Chloro... 170 7e-41
gi28872421 ref NP_795040.1	glutamine synthetase [Pseudomo... 168 2e-40
gi23471345 ref ZP_00126675.1	COG0174: Glutamine synthetas... 168 3e-40
gi32422469 ref XP_331678.1	hypothetical protein [Neurospo... 167 3e-40
gi48764498 ref ZP_00269050.1	COG0174: Glutamine synthetas... 167 4e-40
gi26991088 ref NP_746513.1	glutamine synthetase, putative... 167 4e-40
gi48783610 ref ZP_00280062.1	COG0174: Glutamine synthetas... 167 4e-40
gi26991975 ref NP_747400.1	glutamine synthetase, putative... 167 5e-40
gi15597236 ref NP_250730.1	probable glutamine synthetase ... 166 1e-39
gi23105727 ref ZP_00092181.1	COG0174: Glutamine synthetas... 166 1e-39
gi46164186 ref ZP_00136728.2	COG0174: Glutamine synthetas... 166 1e-39
gi26991859 ref NP_747284.1	glutamine synthetase, putative... 165 2e-39
gi23471983 ref ZP_00127311.1	COG0174: Glutamine synthetas... 165 2e-39
gi15595493 ref NP_248987.1	probable glutamine synthetase ... 165 3e-39

4. I hereby declare that all statement made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these

Applicant : Andrew Kloeck et al.
Serial No. : 10/098,602
Filed : March 15, 2002
Page : 4 of 4

Attorney's Docket No.: 12557-004001

statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application or any patent issued thereon.

Date: 7/14/04

Andrew P. Kloeck
ANDREW P. KLOECK